

Short Communication

Essential Oils Against Foodborne Pathogens and Spoilage Bacteria in Minced Meat

Lidiane Nunes Barbosa,¹ Vera Lucia Mores Rall,¹ Ana Angélica Henrique Fernandes,²
Priscila Ikeda Ushimaru,¹ Isabella da Silva Probst,¹ and Ary Fernandes Jr.¹

Abstract

The antimicrobial activity of essential oils of oregano, thyme, basil, marjoram, lemongrass, ginger, and clove was investigated *in vitro* by agar dilution method and minimal inhibitory concentration (MIC) determination against Gram-positive (*Staphylococcus aureus* and *Listeria monocytogenes*) and Gram-negative strains (*Escherichia coli* and *Salmonella* Enteritidis). MIC_{90%} values were tested against bacterial strains inoculated experimentally in irradiated minced meat and against natural microbiota (aerobic or facultative, mesophilic, and psychrotrophic bacteria) found in minced meat samples. MIC_{90%} values ranged from 0.05%v/v (lemongrass oil) to 0.46%v/v (marjoram oil) to Gram-positive bacteria and from 0.10%v/v (clove oil) to 0.56%v/v (ginger oil) to Gram-negative strains. However, the MIC_{90%} assessed on minced meat inoculated experimentally with foodborne pathogen strains and against natural microbiota of meat did not show the same effectiveness, and 1.3 and 1.0 were the highest log CFU/g reduction values obtained against tested microorganisms.

Introduction

THE NEW TECHNOLOGIES of food preservation include nonthermal inactivation, such as ionization radiation, high hydrostatic pressure, and pulsed electric fields; modified atmosphere and active packaging; biopreservation; and natural antimicrobial compounds (Devlieghere *et al.*, 2004). Plants are a source of bioactive molecules and have been widely used both traditionally and commercially to increase the shelf-life and safety of foods (Sasidharan *et al.*, 2008).

Biological properties of essential oils and their antimicrobial activity have been attributed to phenolic compounds, such as the carvacrol, eugenol (2-methoxy-4-(2-propenyl) phenol), and thymol (Seydim and Sarikus, 2006). These compounds have hydrophobic characteristics and interact with different sites of microbial cell (e.g., cell wall and cytoplasmic membrane), causing loss of cellular constituents, collapse of membrane structure, and cell death (Burt, 2004). Bactericidal or bacteriostatic activity of essential oils, *in vitro* and in food assays, against

Salmonella enterica, *Escherichia coli* O157:H7, *Staphylococcus aureus*, *Listeria monocytogenes*, *Lactobacillus plantarum*, *Saccharomyces cerevisiae*, and *Candida albicans* strains has been reported (Lambert *et al.*, 2001; Chorianopoulos *et al.*, 2004; Friedman *et al.*, 2004; Kim *et al.*, 2004). Studies *in vitro* have used spices as antimicrobials in laboratory media although the levels of spices and their essential oils to inhibit microorganisms in food have been found to be higher than those assays performed using culture media (Burt and Reinders, 2003; Uhart *et al.*, 2006).

Thus, we aimed to determine *in vitro* the minimal inhibitory concentration (MIC) of essential oils from *Thymus vulgaris* (thyme), *Origanum majorana* (marjoram), *Origanum vulgare* (oregano), *Ocimum basilicum* (basil), *Zingiber officinale* (ginger), *Cymbopogon citratus* (lemon grass), and *Caryophyllus aromaticus* (clove) against *E. coli*, *S. aureus*, *L. monocytogenes*, and *Salmonella* Enteritidis strains. MIC_{90%} values were evaluated in minced meat irradiated and experimentally inoculated with these pathogenic bacteria and against natural microbiota of minced meat (mesophiles and psychrotrophs).

Departments of ¹Microbiology and Immunology and ²Chemistry and Biochemistry, Biosciences Institute, São Paulo State University, Sao Paulo, Brazil.

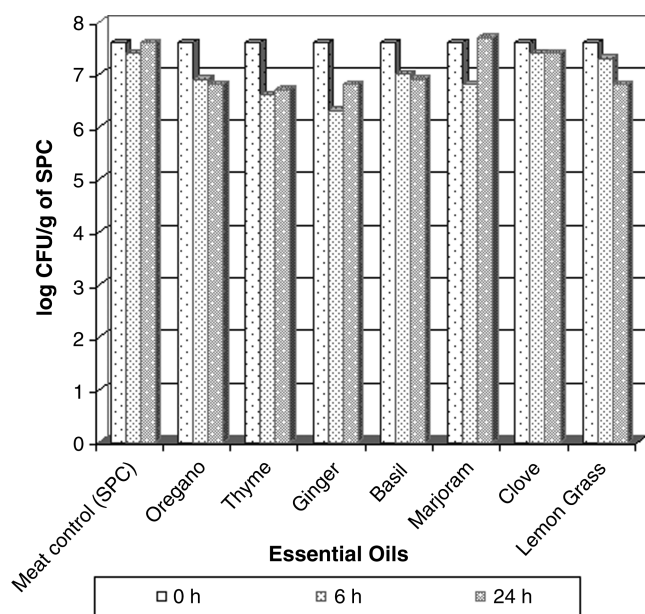


FIG. 2. Log of CFU/g of standard plate count (SPC) values recorded on minced meat samples after 5°C/6 and 24 h of essential oil addition.

oils and clove oil for Gram-negative strains (0.10%v/v). *In vitro* studies have demonstrated the antibacterial activity of essential oils against *L. monocytogenes*, *Salmonella* Typhimurium, *E. coli* O157:H7, *Shigella dysenteriae*, *Bacillus cereus*, and *S. aureus*, and Gram-negative bacteria were less susceptible than Gram-positive bacteria (Burt, 2004).

The CFU/g (log) values of *S. aureus*, *L. monocytogenes*, *E. coli*, and *Salmonella* Enteritidis assays with or without essential oil zacontact in meat experimentally inoculated are shown in

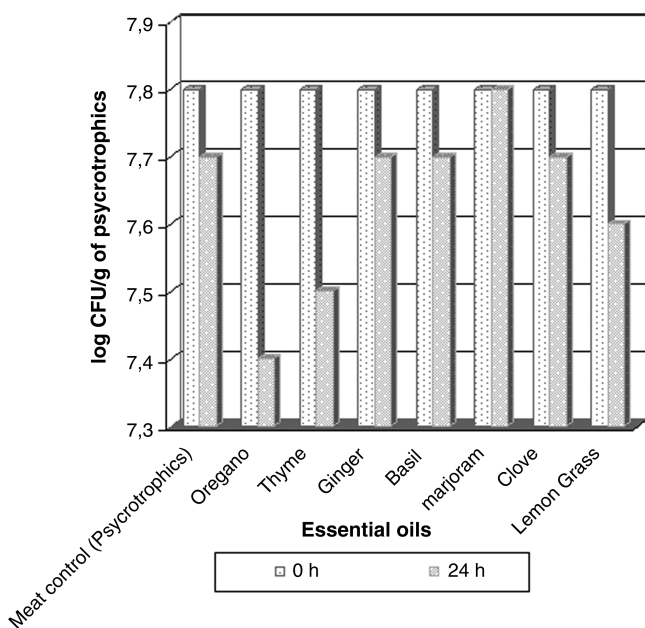


FIG. 3. Log of CFU/g for psychrotrophic microorganism values recorded on minced meat samples after 5°C/24 h of essential oil addition.

Fig. 1. The reduction between tests and control treatments was not significant, and the oils were able to reduce 1 log compared with the control. Although no significant differences were found, the bacteriostatic effect of oils was verified, and no bacterial developments were recorded at 5°C/3 h for all bacterial strains.

The log CFU/g values for mesophilic aerobic bacteria from minced meat recorded at 0 h (positive control) and 6 and 24 h after adding oil to meat are presented in Fig. 2. No significant differences were verified, and 1.3 and 1.0 were log CFU/g reduction values to ginger and thyme oils, respectively. The psychrotrophic reduction tests (Fig. 3) after 24 h of oils and meat contact produced the highest log reduction with oregano oil (0.4).

Although the antimicrobial activity *in vitro* of oils has been moderately effective on meat model, the potential use of these oils in food preservation technologies should be found in optimal concentrations to ensure the safety of the food, appropriated organoleptical characteristics, and accepted by consumers. Studies aiming to elucidate the interaction between essential oils and components of food matrices or additives, stability of oils during food processing, and the standardization of antibacterial methods are still needed.

Acknowledgments

This research was supported by Fundação de Amparo a Pesquisa de São Paulo (FAPESP 05/56110-2 and 05/55039-2). We thank the Companhia Brasileira de Esterilização (CBE) for meat sample irradiation, Dr. Luciano Barbosa for statistical analysis, and Dr. José Maurício Sforcin for critical review of the article.

References

Burt S. Essential oils: their antibacterial properties and potential applications in foods—a review. *Int J Food Microbiol* 2004;94: 233–253.

Burt SA and Reinders RD. Antibacterial activity of selected plant essential oils against *Escherichia coli* O 157:H7. *Lett Appl Microbiol* 2003;36:162–167.

Chorianopoulos N, Kalpoutzakis E, Aliogiannis N, Mitaku S, Nychas G, and Haroutounian SA. Essential oils of *Satureja*, *Origanum*, and *Thymus* species: chemical composition and antibacterial activities against foodborne pathogens. *J Agric Food Chem* 2004;52:8261–8267.

Devlieghere F, Vermeiren L, and Debevere J. New preservation technologies: possibilities and limitations. *Int Dairy J* 2004;14: 273–285.

Friedman M, Henika PR, Levin CE, and Mandrell RE. Antibacterial activities of plant essential oils and their components against *Escherichia coli* O157:H7 and *Salmonella enterica* in apple juice. *J Agric Food Chem* 2004;52:6042–6048.

Kim JW, Kim YS, and Kyung KH. Inhibitory activity of essential oils garlic an onion against bacteria and yeasts. *J Food Prot* 2004;67:499–504.

Lambert RJW, Skandamis PN, Coote PJ, and Nychas GJE. A study of the minimum inhibitory concentration and mode of action of oregano essential oil, thymol and carvacrol. *J Appl Microbiol* 2001;91:453–462.

[NCCLS] National Committee for Clinical Laboratory Standards. *Method for Dilution Antimicrobial Susceptibility Tests for Bacterial that Grow Aerobically*, 7th edition. Approved Standard M7.A6. NCCLS: Wayne, PA, 2004.

- Sasidharan S, Zuraini Z, Yoga Latha L, Sngetha S, and Suryani S. Antimicrobial activities of *Psophocarpus tetragonolobus* (L.) DC extracts. *Food Pathog Dis* 2008;5:303–309.
- Seydim AC and Sarikus G. Antimicrobial activity of whey protein based edible films incorporated with oregano, rosemary and garlic essential oils. *Food Res Int* 2006;39: 639–644.
- Uhart M, Maks N, and Ravishankar S. Effect of spices on growth and survival of *Salmonella* Typhimurium DT 104 in ground beef stored at 4 and 8°C. *J Food Saf* 2006;26:115–125.

Address correspondence to:
Ary Fernandes Jr., Ph.D.
Department of Microbiology and Immunology
Biosciences Institute
São Paulo State University
IBB/UNESP/Botucatu
Sao Paulo 18618-000
Brazil
E-mail: ary@ibb.unesp.br